Viability and histological appearance of minced cartilage and freshly isolated chondrons from human osteochondral allografts

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ABSTRACT

INTRODUCTION: Articular cartilage has no ability for self-repair. Therefore, cartilage injury often leads to osteoarthritis. Cartilage repair methods such as osteochondral allografts and autologous chondrocyte implantation can restore cartilage function and decrease complaints. However, these treatments are resource intensive andlogistically challenging, and thus low-cost alternatives like minced cartilage implantation are emerging in the clinic. Fundamental understanding of this technique is currently limited. Arthroscopically minced cartilage is often called ‘chondron’ which is an isolated cartilage cell surrounded by pericellular matrix composed of type VI collagen. The objective of this study is to compare isolated chondrocytes and chondrons, minced cartilage and OCA in terms of cell viability and histological appearance.

METHODS: Cartilage was procured from leftovers of fresh human OCAs. Grafts were immediately processed for experiments. The groups evaluated were: 4mm biopsies (control, group A), arthroscopically minced cartilage (using an arthroscopic shaver and AutoGraft collector, group B), cartilage minced by hand (group C), isolated chondrons (digested for overnight with 0.75 units/mL dispase and 125 U/mL collagenase type 2, group D) or chondrocytes (group E, 500 U/mL collagenase type 2). Biopsies, minced cartilage, or cells were embedded in fibrin gels. Viability was assessed using fluorescence microscopy after staining with Calcein AM and ethidium homodimer-1. Viability was quantified as percentage of total cells using ImageJ. Gels were embedded in paraffin and stained using safranin-O and immunohistochemistry for type II and VI collagen.

RESULTS: Average viability of osteochondral allografts was 46.3±1.9%. Isolated chondrocytes had a higher viability with 67.4±9.9% compared to biopsy controls and arthroscopically minced cartilage (Figure 1). Safranin-O staining and collagen type II is positive for proteoglycans in biopsies and minced cartilage but not in uncultured chondrons and chondrocytes (Figure 2). Collagen type VI is present in the pericellular matrix in tissues samples and isolated cells.

DISCUSSION: Relative viability in arthroscopically minced cartilage is lower compared to isolated chondrons and chondrocytes and contains a surrounding extracellular matrix staining positive for proteoglycans. The term chondron and minced cartilage cannot be used interchangeably. Freshly isolated chondrocytes also contain type VI collagen in their pericellular matrix, indicating that current concentration of collagenase type 2 does not rigorously remove type VI collagen matrix.

SIGNIFICANCE/CLINICAL RELEVANCE: This study shows that minced cartilage and chondrons have important differences in terms of presence of extracellular matrix and viability. The clinical consequences of these differences remain to be elucidated.

![Figure 1. Viability of minced cartilage and isolated cells.](image1.png)
Each point represents a donor and 6 replicates per donor.

![Figure 2. Histologic appearance.](image2.png)
Safranin-O and type II collagen staining are positive in allograft tissue biopsy and arthroscopically and hand minced cartilage surrounded by fibrin glue, but not in isolated chondrons and chondrocytes. All groups show pericellular location of type VI collagen.